Anisakidae nematodes and Trypanorhyncha cestodes of hygienic importance infecting the king mackerel *Scomberomorus cavalla* (Osteichthyes: Scombridae) in Brazil

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From February to October 2007, thirty specimens of the king mackerel, *Scomberomorus cavalla* (Cuvier, 1829) were purchased from markets in the municipalities of Niterói and Rio de Janeiro. The fishes were measured, filleted and further had their organs investigated for helminths. Ten out of the thirty fish specimens were parasitized with anisakid nematodes represented by *Anisakis* sp. and *Contracaecum* sp. with prevalence of 1% and 16%, mean intensity of 2 and 3.31 and mean abundance of 0.02 and 0.53, respectively. The infection range with *Contracaecum* sp. was 1–9. The sites of infection were the stomach serosa and mesentery. Seventeen fish specimens (53%) out of the 30 investigated were parasitized with Trypanorhyncha metacestodes, identified as *Callitetrarhynchus gracilis*, *Pterobothrium crassicole*, *Callitetrarhynchus speciosus* and *Tentacularia coryphaenae* in the mesentery, with prevalence of 26, 20, 6, 3%, intensity and mean intensity of 3.25, 3.5, 1, 2 and mean abundance of 0.86, 0.7, 0.06 and 0.06, respectively. The infection range due to *C. gracilis* and *P. crassicole* were of 1–5 and 1–20, respectively. *Anisakis* sp., *C. speciosus* and *P. crassicole* are reported in *S. cavalla* for the first time. Considerations on the zoonotic potential of the parasites and their rules in sanitary inspection are presented.

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1. Introduction

The king mackerel, *Scomberomorus cavalla* (Cuvier, 1829) occurs in the occidental Atlantic, from Massachusetts, in the USA, to the south of Brazil. Basically, specimens of this species fed on other fishes, mollusks and crustaceans, mainly squids and shrimps (Figueiredo and Menezes, 2000).

In Brazil, data on the fishery of this species indicate its high economic value related to the amount of fish obtained, taking into account the evaluated expressive internal acceptance of the product; moreover meat of the king mackerel has been exported to the USA, France, Spain, Argentina, Portugal and Japan (IBAMA, 2007). The species is still poorly studied considering hygienic-sanitary procedures regarding ichthyoparasitological approaches. Among the possible harbored parasites by specimens of the king mackerel, two groups are of concern: the Anisakidae nematodes, with zoonotic potential and the Trypanorhyncha cestodes, due to their importance during sanitary inspection, if one considers the repulsive aspect of infected meat certainly causes to consumers.

The presence of parasites in fish products indicates a harmful sanitary problem not to be underestimated. Even considering that most of the parasitic agents is not pathogenic to humans, some species can be associated to serious diseases due to the ingestion of contaminated...
fish as occurs, as widely known, in those caused by larval anisakid nematodes (González, 2003).

Human infections with those parasites can settle after the ingestion of either raw or poorly processed fish meat thus preserving the larval viability. Historically, those infections were most frequent in localities where typical alimentary habits are adopted. Nevertheless, presently, some of these infections are of surpassing medical concern in several countries, due to the increasing popularity of some oriental foods and to the encouraging appeals of naturalists that recommend the ingestion of either raw or poorly cooked fishes (Ferre, 2001).

To prevent the infection is necessary that people avoid ingesting fish meat either prepared under these conditions or inadequately processed during cooking, freezing, salting, smoking procedures or try to inactivate the parasites before the commercialization of infected fishes. In accordance with the guidelines of the North American agency “Food and Drug Administration” (FDA, 2003), fish meat bound to ingestion and prepared under temperatures below 60 °C has either to be frozen under −35 °C for, at least, 15 h or under −20 °C for seven days. The visual analysis of contaminated specimens to be further discharged is recommended and foreseen by the legislation of several countries including Brazil, Spain and France, among others (Brasil, 2007). In industries and in this type of analysis, the candling table can be utilized in the samplings of filleted meat.

According to Smith and Wootten (1978) fish eversion immediately after capture is a control procedure that prevents larval migration from the viscera to the musculature; nevertheless, viscera are not to be discharged in water collections. Also some authors suggest that the hypoaxial musculature may be eliminated considering its high worm burdens, aiming at the reduction of potential risks related to anisakiasis (Herreras et al., 2000).

Since the detection of the parasitism and associated lesions in fishes infected with anisakid larvae and the appearance of the symptoms in humans, this parasitosis has been considered as emerging and a target of investigations aiming at the development of techniques to support the safe alimentary production as well as reliable methodologies for laboratorial diagnosis, besides the adoption of adequate preventive procedures. In several countries studies aim at the knowledge of hosts, infection dynamics, the relation between fish parasites and the resulting problems of public health importance. Records of anisakid larvae parasitizing marine fishes from the littoral of Rio de Janeiro have been made by several authors (São Clemente et al., 2007). The species Tentacularia coryphaenae Bosc, 1802 and Callitetrarhynchus gracilis (Rudolphi, 1819) Pintner, 1931, although not reported from S. cavalla in Brazil, were referred in other scombrid fishes (Alves and Luque, 2006). Overseas, trypanorhynchs were recovered from specimens of S. cavalla, in Miami, USA, Ward (1954) in Battes (1990) reported to the presence of T. coryphaenae, and Palm and Overstreet (2000) referred to C. gracilis in the Gulf of Mexico.

In accordance with Dollfus (1942) cysts of Trypanorhyncha are not transmissible to homeothermic vertebrates and the re-encapsulation of the post-larvae does not occur in this group of hosts; nevertheless, according to Deardorff et al. (1984) the localization of larvae in the musculature of fishes can release toxins and, consequently, affect humans. Some reports have demonstrated that extracts from a species of Trypanorhyncha are responsible for immune responses in mice, indicating the possibility of allergic reactions in humans (Rodero and Cuéllar, 1999; Vázquez-López et al., 2001, 2002; Gómez-Morales et al., 2008).

The present paper aimed the study of the parasitism of larval Anisakidae and Trypanorhyncha worms infecting the king mackerel S. cavalla (Cuvier, 1829), commercialized in the State of Rio de Janeiro, Brazil, with data on the helminth species, parasitological indexes related to prevalence, mean intensity, mean abundance, infection range, sites of infection and sanitary aspects due to their importance in fish hygiene and public health programs.

2. Materials and methods

From February to October 2007, 30 specimens of S. cavalla (Cuvier, 1829) with total lengths of 81.1–141 cm
Thirty-three percent of the 30 purchased specimens of *S. cavia* were infected with third-stage Anisakidae larvae, representing the species *Anisakis* sp. and *Contracaecum* sp.; 53% with the Trypanorhynchia cestodes: *T. coryphaenae*, *Pterobothrium crassicole* Diesing 1850, *C. gracilis* and *Callitetrarhynchus speciosus* (Linton, 1897) (Carvajal & Rego, 1985).

Parasitological indexes related to prevalence, intensity and mean intensity, infection range, mean abundance, as well as the infection sites and CHIOC deposit numbers are depicted in Table 1.

### Table 1

<table>
<thead>
<tr>
<th>Helminth species</th>
<th>P (%)</th>
<th>I'/MI</th>
<th>MA</th>
<th>IR</th>
<th>Infection site</th>
<th>CHIOC no.</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Anisakis</em> sp.</td>
<td>1</td>
<td>2</td>
<td>0.02</td>
<td>–</td>
<td>Stomach serosa</td>
<td>35634</td>
</tr>
<tr>
<td><em>Contracaecum</em> sp.</td>
<td>16</td>
<td>3.31</td>
<td>0.53</td>
<td>1–9</td>
<td>Mesentery</td>
<td>35635</td>
</tr>
<tr>
<td><em>Callitetrarhynchus gracilis</em></td>
<td>26</td>
<td>3.25</td>
<td>0.86</td>
<td>1–5</td>
<td>Mesentery</td>
<td>35630</td>
</tr>
<tr>
<td><em>Pterobothrium crassicole</em></td>
<td>20</td>
<td>3.5</td>
<td>0.7</td>
<td>1–20</td>
<td>Mesentery</td>
<td>35632</td>
</tr>
<tr>
<td><em>Callitetrarhynchus speciosus</em></td>
<td>6</td>
<td>1</td>
<td>0.06</td>
<td>–</td>
<td>Mesentery</td>
<td>35631</td>
</tr>
<tr>
<td><em>Tentacularia coryphaenae</em></td>
<td>3</td>
<td>2</td>
<td>0.06</td>
<td>–</td>
<td>Mesentery</td>
<td>35633</td>
</tr>
</tbody>
</table>

* Intensity is indicated only when a host was collected.

(110.5 cm) were purchased from fish markets in the municipalities of Niterói and Rio de Janeiro, State of Rio de Janeiro, Brazil. Specimens were identified in accordance with Figueiredo and Menezes (2000). Fishes were preserved in isothermal boxes with ice, to be further transferred to the Laboratory of Inspection and Fishery Technology of the Veterinary School, Fluminense Federal University, where specimens were identified, measured, submitted to necropsy and filleted. Helminths were recovered in Petri dishes with a 0.65% NaCl solution. Nematodes were fixed with hot (60°C) AFA, preserved in 70% GL ethanol–glycerin, clarified with Aman’s lactophenol. Plerocercoids and plerocerci of Trypanorhynchia were transferred to distilled water, where cysts of plerocerci were broke open with the aid of sharp needles under the stereomicroscope to release the larvae that were put in the refrigerator for at least 24 h to permit the relaxing of scolices and tentacular extroversion. Further, larvae were fixed in cold AFA, stained with Langeron’s carmine, differentiated in a 5% chloridric ethanol solution, dehydrated in a crescent alcoholic series, clarified in beechwood creosote and preserved either as whole mounts in Canada Balsam or in 70% GL ethanol. Taxonomic generic identification of larval nematodes was based on Rego et al. (1983) and Petter and Maillard (1988); Trypanorhynchia cestodes were identified in accordance with Carvajal and Rego (1985), Campbell and Beveridge (1994, 1996). Indexes of prevalence, intensity, mean intensity, mean abundance and infection range follow Bush et al. (1997). Representative parasite specimens were deposited in the Helminthological Collection of the Oswaldo Cruz Institute (CHIOC), Rio de Janeiro, Brazil.

Anisakis Dujardin, 1845. *Anisakis* sp. Main morphological characteristics based on two third-stage larvae: cuticle with thin transversal striation. Anterior extremity with a dorsal and two poorly developed ventro-lateral lips. Six cephalic papillae, one pair in the dorsal lip and a pair in each ventro-lateral lip. Boring tooth below the oral aperture, between the two ventro-lateral lips. Excretory pore opening beneath the boring tooth. Deirids inconspicuous. Ventriculus longer than large. Ventricular appendix and intestinal cecum absent. Two nearly spherical rectal glands. Tail conical, mucron present.

*Contracaecum* Railliet & Henry, 1912. *Contracaecum* sp. Main morphological characteristics based on ten third-stage larvae: cuticle with thin transversal striation, more evident in the posterior extremity of the body. Anterior extremity with a dorsal lip and two poorly developed ventro-lateral lips. Six cephalic papillae, one pair in the dorsal lip and a pair in each ventro-lateral lip. Boring tooth near the oral aperture, between the two ventro-lateral lips. Excretory pore opening beneath the boring tooth. Deirids inconspicuous. Ventriculus small and sub spherical. Ventricular appendix nearly twice longer than the intestinal cecum. Two almost spherical rectal glands. Tail conical, mucron absent.


3. Results

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present. Hook files 1(1') widely separated, falciform; intercalary row(s) present proximal to each principal row; intercalary rows extend onto external surface to merge with band of hooks occupying midline of external surface of tentacle. Sheaths very sinuous. Bulbs elongate. Pars postbulbosa present.

_Callitetrarhynchus Pintner, 1931: Callitetrarhynchus gracilis_ (Rudolphi, 1819) Pintner, 1931. Main morphological characteristics based on ten plerocerci: plerocercus with blastocist, with caudal extension. Scolex long, slender, feebly craspedote. Two patelliform bothria, notched on posterior margin. Pars vaginals long, tentacle sheaths regularly sinuous, enlarged anteriorly. Numerous gland cells surround tentacle sheaths from bulbs through most of pars vaginals. Bulbs relatively short. Retractor muscles originate in anterior third of bulb. Pars postbulbosa absent. Armature poecilocanthis, without basal armature or enlargement. Principal rows half-spiral, alternate, hooks, 6(6'), 7(7'), 8(8') form distinct triad separate from chainette; the satellite hooks 7(7') are almost twice longer than hooks 8(8'). Intercalary rows absent. Chainette simple, bases of elements without wings, consisting of hooks 9(9') well separate and forming single file on middle of external face.

_Callitetrarhynchus speciosus_ (Linton, 1897). _Carvajal and Rego_ (1985). Main morphological characteristics based on two plerocerci: plerocercus has a longer scolex and different size and proportions of the scolex parts, almost twice than observed in specimens of _C. gracilis_.

The following morphological differences enabled us to separate the two species. _C. speciosus_ has a longer scolex and different size and proportions of the scolex parts. Despite basic similarities in the arrangement and measurements of hooks, there are many differences in the oncotaxy. Hooks 1(1') in _C. speciosus_ are arranged in a parallel pattern without having their points convergent as in _C. gracilis_; hooks 2(2') are much longer in _C. speciosus_; hooks 3(3') are also larger and have bigger bases. All other hooks, with the exception of 5(5') and 7(7') are larger in _C. speciosus_. The satellite hooks 7(7') and 8(8') are different: in _C. speciosus_ they are almost equal in size, throughout the length of the tentacle but in _C. gracilis_ the satellite hooks 7(7') are almost twice longer than hooks 8(8') along much of tentacle. Furthermore, the chainette is distinct in two species, the hooks being sunken into the tegument in _C. speciosus_, whereas they are higher and have wider bases in _C. gracilis_. Additional differences were noted in the frontal glands which are widespread in _C. speciosus_ and extend from the posterior border of the bothria to the posterior border of pars postbulbosa, whereas in _C. gracilis_ the glands do not extend into anterior quarter of the pars vaginals.

This is the first report of _Anisakis sp._, _Contracaecum sp._, _T. coryphaenae_, _P. crassicole_, _C. gracilis_ and _C. speciosus_ in _S. cavalla_ from Brazil.

4. Discussion

The fact that anisakid larvae were found in the stomach serosa and/or mesentery of fishes represents a low risk of ingestion by consumers. However, the possibility of larval migration to the musculature, while fishes remain uneviscerated on board and/or in warehouses has always to be taken into account. Parasitological surveys of several fish species captured off the Brazilian coast have been reported aiming at a proper understanding of the role larval fish parasites play and their zoonotic potential, in order to supply data so that sanitarins can better detect the parasites and prevent an officially undescribed disease in our country so far (Tavares and Luque, 2006; Knoff et al., 2007).

The Trypanorhynchus recovered from specimens of _S. cavalla_ were represented by _T. coryphaenae_, _C. gracilis_, _C. speciosus_ and _P. crassicole_, the two latter referred for the first time in Brazilian scombrids. It was observed that these cestode species occur in several teleosteans of commercial importance in Brazil (São Clemente, 1986; São Clemente et al., 1995, 2004, 2007; Rego, 1987; Amato et al., 1990; Silva et al., 2000; Alves and Luque, 2006; Dias et al., 2009).

5. Conclusion

It must to been reinforced the necessity to better inform those in charge of the fish hygienic-sanitary control as well as the staffs dealing with mass production activities, from capture to final consumers about the ichthyoparasitoses and prophylactic approaches with the improvement of sanitary educational programs reaching different levels.

It is suggested the adoption of the Hazard Analysis and Critical Control Point (HACCP) during the steps of mass production, aiming the elimination, avoidance or risks reduction, in order to provide a qualified and safe final product and in this case, with fish evisceration on board, warehouses, industries or retail dealers with appropriate discharges.

Multidisciplinary researches are to be developed in the evaluation symptoms in patients with gastroenteric lesions and/or allergic reactions suggesting anisakiasis and their relation with the ingestion or manipulation of raw fish; also, investigations of Trypanorhyncha cestodes are to be carried out in order to evaluate their zoonotic potential considering the association between their released toxins and the settlement of allergies in humans.

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References


